molecules, which then act in competition for the activation sites of the operons or regulons under quorum sensing control. Control of pathogenicity in this way may offer a unique form of antimicrobial therapy without the endotoxemia associated with Gram-negative cell lysis.

S63 A new approach for controlling bacterial populations (sonic communication with bacteria)

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Introduction and objectives: We previously reported that bacteria communicate with each other by using growth-regulating sonic/ultrasonic signals (Matsuhashi et al, J Gen Appl Microbiol 1996; 42: 315–323). *Bacillus subtilis* cells emit sounds at frequencies of about 8.5 kHz and its overtones, and the growth of *B. carboniphilus* cells is promoted by sounds/ultrasounds from a speaker at similar frequencies under non-permissive stress conditions (Matsuhashi et al, J Gen Appl Microbiol 1998; 44: 49–55). Graphite, charcoal and other materials convert external energy, such as pulses of infrared light, into sonic signals (Matsuhashi et al, J Gen Appl Microbiol 1997; 43: 225–230). The response of bacteria to sounds may be utilized in antibiotic therapy.

Results: (1) Soft solid matter, such as solidified agar in growth media, can significantly promote or inhibit the growth of bacteria and yeast under stress conditions and the hatching of fish eggs. The effect is probably due to photo-acoustic emission resulting from the conversion of external electromagnetic energy and can be diminished by shielding the agar and organisms with aluminum foil. *E. coli* and yeasts also responded to signals from the agar. This mechanism probably explains why many bacteria and mycoplasmas grow well on soft solid surfaces such as animal tissue. (2) In contrast to *B. carboniphilus*, soft low-frequency sounds from a speaker, at frequencies between 10 and 800 Hz, markedly inhibit the growth of *E. coli*. This *E. coli* behavior, which may be common in related Gram-negative bacilli, suggests that low-frequency sounds may help to overcome infections by Gram-negative bacteria.

S64 Quorum sensing—a new target for antibiotic therapy

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There are two broad strategies for controlling bacterial infection: either (1) kill the organism or (2) attenuate virulence such that the infecting oragnism fails to adapt to the host environment and can be cleared by the innate host defenses. The latter approach has, until recently, lacked specific targets for rational drug design. However, the discovery that bacterial cells communicate with each other using small diffusible signaling molecules to regulate virulence in concert with cell population density (termed quorum sensing) now offers such a novel target.

Many Gram-negative bacteria employ *N*-acylhomoserine lactones (AHLs) as quorum sensing signaling molecules which not only control gene expression but also possess potent pharmacologic activities such that they may function as virulence determinants *per se*. Understanding the molecular mechanisms by which AHLs are produced (usually via a member of the Lux family of AHL synthases) and by which AHLs activate target gene expression (via members of the LuxR family of transcriptional regulators) is central to the design of small-molecule antagonists (Quorum Sensing Blockers, QSBs) capable of attenuating virulence through the blockade of either signal generation or signal transduction.

Toxicity of antimicrobial agents: relevance of preclinical data for the clinician

S67 Immunologic reactions to antimicrobial agents

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This presentation reviews the incidence, clinical manifestations, differential diagnosis, risk factors and pathogenesis of allergic reactions of two important classes of antimicrobials; beta-lactams and sulfonamides. The diagnostic work-up of a patient with a history of an allergic reaction will be discussed, as well as the possibility of the safe administration of the drug in the face of an allergy using immunotherapy. Emerging concepts of beta-lactam side-chain allergy, the role of cellular immune mechanisms and the clinical importance of cross-reactivity of allergic reactions to different classes of beta-lactams will be emphasized.

S68 Toxic effects of antimicrobial agents on the musculoskeletal system

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Several groups of antimicrobial agents have the potential to have adverse effects on the musculoskeletal system. At the beginning of the 1960s, the effects of tetracycline on the developing skeleton were investigated in premature infants by multiple X-ray examinations. Based on these results, tetracylines were considered to be contra-indicated in infants and children. Quinolones are another group of frequently used drugs that have toxic effects on connective tissue structures. Due to their cartilage toxicity—as shown in immature dogs and other animals—they are contraindicated in children and adolescents. Human experience with pefloxacin in children showed an unacceptably high incidence of joint toxicity (10%). However, with nalidixic acid, norfloxacin and ciprofloxacin, favorable clinical results in juvenile patients have been published. Also, quinolones have the potential to induce tendon disorders (tendinitis, Achilles tendon rupture) which have occurred mainly in elderly patients. Mechanistic studies from our group indicate that the magnesium-chelating properties of the quinolones might be responsible for these toxic effects. Less often used antimicrobial drugs which might affect the musculoskeletal system include, for example, pyrazinamide, rifabutin, and quinupristin–dalfopristin.

Infectious disease problems in Central and Eastern Europe

S71 Typical problems of infectious diseases in Central Europe

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Central and eastern European countries face the same types of

problems concerning infectious diseases as the majority of the other European countries, but these problems can be much more pronounced in particular countries, depending on the local conditions.

The abuse of antimicrobials is especially heavy in the outpatient setting. This problem is aggravated by the rapid increase in the number of registered antibiotics since the early 1990s, which almost inevitably results in the incorrect use of them.

The bacterial resistance rates vary from country to country; in Hungary they are close to those published in Spain. The correct figures are unknown due to the lack of large-scale, standardized surveillance programs, and the small studies reflect only the local situation. The most important problem bacteria are the penicillin-resistant pneumococci, in some institutions MRSA/MRSE, and the multidrug-resistant Gram-negative rods. The VRE do not pose a problem in this region.

Shortage of money induces attempts to decrease the share of antimicrobials in the hospital budget. These attempts, however, consider only the purchase price of antimicrobials and urge the use of cheaper ones. The restrictions do not result in the rational use of antibiotics, but rather in the suboptimal treatment of patients.

There is no modern infection control in the majority of hospitals.

The influence of infectologists/clinical microbiologists on the daily practice of medical doctors is fairly poor.

S72 Molecular epidemiology of MRSA in Poland

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Methicillin-resistant *Staphylococcus aureus* (MRSA) was reported in Poland in the early 1960s, long before this drug was introduced in the country. However, the first studies on their spread and prevalence started in the 1990s, when no single hospital was free from MRSA. At the same time, a several genotypic methods were introduced into the epidemiologic investigations on MRSA. Over 600 MRSA strains collected in hospitals scattered over the whole country were typed and analyzed by some of these methods. The first was pulsed-field gel electrophoresis of *Sma*I-digested DNA, followed by arbitrary primed PCR (AP-PCR), and randomly amplified polymorphic DNA (RAPD) analysis, restriction fragment length polymorphism (RFLP) analysis of plasmid DNA, and of selected genes (e.g. *spa*, *coa*), analysis of *mecA* polymorphs and Tn*554* insertion. Studies from 1990–92 revealed the spread of two different clusters of MRSA strains. Isolates of the first cluster were always heterogeneously resistant to methicillin and usually susceptible to almost all other antistaphylococcal drugs with the exception of tetracycline. MRSA strains of the second cluster expressed homogeneous resistance to methicillin and were always multidrug resistant. Since that time, spread of these two clusters within the country has been analyzed. This has resulted in identification of the first outbreak of mupirocin-resistant MRSA in 1994 and its intra-hospital spread during the following years. A new epidemic of MRSA in the northeast of Poland in 1996 and an epidemic of MRSA with a defect of clumping factor production were also identified. Phenotypic and genotypic studies on a large collection of MRSA strains from Poland resulted in characterization of Polish MRSA strains and a better understanding of their epidemiology.

S73 Meningococcal invasive disease in central Europe

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Objectives: To assess the influence of ET-15/37 clone on invasive meningococcal disease in some European countries.

Methods: The laboratory data presented at the meetings of the European Monitoring Group on Meningococci were analyzed. These data have been collected since 1993 from the Czech Republic, Slovakia, Poland, Austria and Germany. They were compared with the data analyzed in 1987. The selected *Neisseria meningitidis* strains identified in the countries mentioned above were referred to the National Reference Laboratory for Meningococcal Infections in Prague for multilocus enzyme electrophoresis (MLEE) and pulsed-field gel electrophoresis (PFGE).

Results: In the Czech Republic, meningococcal invasive disease occurred sporadically in the past. This situation changed dramatically in 1993 when a new meningococcal clone was detected. This clone was identified by MLEE as clone ET-15, belonging to the 37 complex, with the prevailing phenotype C:2a:P1.2,P1.5 (80%). The incidence and fatality rate of the disease increased, particularly in the age group 15–19 years. In Slovakia, this clone was found in 1995, causing a similar change in the epidemiologicl and clinical situation. In Austria, Germany and Poland, *Neisseria meningitidis* strains belonging to the ET-15/37 complex were found recently; however, they remain infrequent and the total incidence of invasive meningococcal disease has not increased. PFGE of selected strains revealed differences between the strains isolated in different countries.

Conclusions: In some European countries, *Neisseria meningitidis* strains belonging to the ET-15/37 clone caused invasive meningococcal disease requiring special therapeutic and preventive measures.

S74 Antibiotic policy in Russia

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During the last 2 years, substantial progress has been made in the issues related to antibiotic policy in Russia. This became possible because active implementation of formulary system in the hospitals has been initiated by Ministry of Health, and the efforts of public organizations such as the Commission on Antibiotic Policy of Ministry of Health, the Inter-regional Association on Clinical Microbiology and Antimicrobial Chemotherapy (IACMAC), etc. Also, a lot of attention is paid to the antibiotic policy questions at the biggest Russian annual congresses on humans and medicine. Very good educational support for rational antibiotic policy has come from the ESCMID, ISC, ASM, FESCI and APUA. Another positive driving force has come from the pharmaceutical industry and microbiological companies at local, regional and federal levels. In addition, multicenter, well-designed microbiological in vitro studies have been conducted, and their results were taken into consideration during the development of local antibiotic formularies. However, there are still some specific problems in the implementation of rational antibiotic policy. There is no well-established system of local monitoring of resistance in hospitals, so there is a weak influence of these results on updating of antibiotic formularies. Economic reasons (the so-called barter economy) significantly limit the ability of hospitals to buy antibiotics of their choice. Also, the bad marketing practices (e.g. use of data of foreign pharmacoeconomic studies for marketing purposes, advertisements in the mass media, etc.) of some pharmaceutical companies aggravate the above-mentioned problems. But the most important issue which has not been completely resolved in Russia is

the implementation of the ideology of antibiotic policy, stressing the unique role of antibiotics.

Pathogenesis and prevention of polymer-associated staphylococcal infections

S76 Overview of the clinical presentation and problems of polymer-associated staphylococcal infections, with special emphasis on catheter-related infections

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Polymer-associated infections of staphylococci are, in the majority of cases, caused by coagulase-negative staphylococcl (CNS) and include infections associated with a wide variety of indwelling devices, from intravascular catheters to joint and cardiac valve prostheses to cerebrospinal fluid shunts and devices used for peritoneal dialysis. Infection is one of the commonest and most serious complications of indwelling devices. The clinical presentation varies greatly, and diagnosis may either be straightforward or extremely difficult. Usually there is an indolent clinical picture with low-grade fever and minimal local signs of inflammation, e.g. in cases of intravascular catheter-related infections or prosthetic joint infections. However, in some cases of prosthetic valve endocarditis, there may be a life-threatening course characterized by a sudden onset with high fever and chills and severe dysfunction of the prosthetic valve. The difficulties in diagnosis of the usual case with an indolent course with minimal clinical signs are particularly well illustrated by many cases of intravascular catheter-related infection. This had led to numerous attempts to provide accurate clinical and microbiological criteria to establish the diagnosis of catheter-related infection, with more or less success. Some criteria are, for obvious reasons, not easily applicable to certain patient categories e.g. multiple blood cultures in premature neonates. Others, notably some of the proposed microbiological criteria, require methods that are too cumbersome and time-consuming for a routine clinical laboratory.

A keen clinical eye remains, more often than not, the most helpful instrument for the initial diagnosis; the published clinical criteria may aid, together with some of the more advanced microbiological methods and scanning techniques, in finally establishing the diagnosis of a device-related infection.

S77 Recent insights into molecular pathogenesis

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Staphylococci may colonize foreign material by any of three routes: (1) prior to insertion through the surgical wound; (2) after insertion through catheter hubs or skin wounds; or (3) hematogeneously. Consequently, for initial adhesion to the foreign surface, staphylococci have to interact either with native or with host factor-adsorbed polymer. Both *Staphylococcus aureus* and coagulase-negative staphylococci (CNS) have been shown to avidly interact with proteinaceous host factors such as fibronectin and fibrinogen. A number of bacterial adhesins have been implicated in this adhesion process, and the role of some of these adhesins has been molecularly characterized. In addition to plasma factors, *S. aureus* binds to surface-adsorbed blood platelets and their membrane-exposed granule contents such as thrombospondin or von Willebrand factor (vWF). Molecular analysis indicates a role of two fibrinogen-binding adhesins, i.e. Coa/FbpA and Efb, in platelet binding, and of protein A in vWF binding. Adhesion of CNS to native polymer is mediated by an autolysin, AtlE, which influences bacterial surface characteristics and binds to plasma proteins such as vitronectin. After initial adhesion, staphylococci may accumulate on the surface, resulting in biofilm formation. Products of the recently identified *icaADBC* gene cluster in CNS confer production of polymerized polysaccharide antigens such as PIA and PS/A, are involved in phase variation and are associated with clinical disease. Furthermore, biofilm formation requires expression of an accumulation-associated protein, AAP. In summary, a number of molecularly characterized mechanisms contribute to the ability of staphylococci to colonize and infect foreign material. Future research activities may be directed towards enhanced understanding of the respective role and regulation of these mechanisms under the complex conditions present in vivo.

S78 Lessons learned from animal models and from use in humans

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Microbial adhesion to foreign material is an initial and fundamental mechanism for the development of infection. On their surfaces, staphylococci, in particular *Staphylococcus aureus*, express several receptors or adhesins which interact with specific host proteins such as fibrinogen, fibronectin, collagen, vitronectin, laminin, thrombospondin, bone sialoprotein, or elastin. Information on the molecular structure of *S. aureus* genes coding for bacterial adhesins is growing rapidly. Cloning, sequencing and site-directed mutagenesis of the corresponding genes for major *S. aureus* adhesins have allowed the characterization of the molecular structure and function of one fibrinogen-binding protein (=clumping factor), one collagen adhesin, and two distinct but related fibronectin-binding proteins.

Site-specific mutants of each individual adhesin showed specific defects in adhesion to their respective host proteins. Furthermore, adhesion-defective mutants complemented with functional genes (located either on multicopy plasmids or integrated into the bacterial chromosome) allowed full restoration of each adhesion phenotype. The ligand-binding domains of either fibronectin, fibrinogen or collagen adhesins have been identified by binding and inhibition studies performed with recombinant truncated protein fragments or synthetic peptides. Animal models of endocarditis and bioimplants have shown that mutants of *S. aureus* defective in either a fibronectin or a fibrinogen adhesin have less ability to attach and to induce experimental infection. Elucidation of molecular mechanisms of bacterial attachment to host tissues and biomedical implants as well as invasion of non-phagocytic cells may lead to the development of better therapeutic approaches for *S. aureus* infections.

Clinical impact of new diagnostic methods in microbiology

S80 New automated systems for identification and antibiotic resistance detection

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Automation of the bacteriology laboratory has not yet become a reality. However, new instruments such as the VITEK 2 have recently